

## Patent claims 1 to 21 according to Article 34 PCT

1. Set of nucleic acid molecules by means of which, in a process for the detection of representatives of *Salmonella enterica* subsp. *enterica*, *salamae*, *arizonae*, *diarizonae*, *houtenae*, *bongori* and *indica*, all the representatives of those subspecies can be detected, which set of nucleic acid molecules is **obtainable** by

(a) obtaining or deriving a first nucleic acid molecule (nucleic acid molecule 1) in a manner known *per se* using a nucleic acid isolate of a representative of one of the mentioned *Salmonella enterica* subspecies, which first nucleic acid molecule is specifically suitable as primer or probe for the detection of that representative or of further or all representatives of that one *Salmonella enterica* subspecies and possibly also of representatives of further *Salmonella enterica* subspecies,

(b) obtaining or deriving a second nucleic acid molecule (nucleic acid molecule 2) in a manner known *per se* using a nucleic acid isolate of a different representative of one of the mentioned *Salmonella enterica* subspecies, which second nucleic acid molecule is specifically suitable as primer or probe for the detection of that representative or of further or all representatives of that different *Salmonella enterica* subspecies and possibly also of representatives of others of the mentioned *Salmonella enterica* subspecies, and

(c) unless it is already possible to detect all the representatives of the mentioned *Salmonella enterica* subspecies using the nucleic acid molecules obtainable or

derivable according to (a) and (b), continuing to obtain or derive nucleic acid molecules according to (a) and/or (b) until all the representatives of the mentioned *Salmonella enterica* subspecies can be detected using the obtained or derived set of nucleic acid molecules,

wherein

(d) the nucleic acid isolates comprise or are phylogenetically conserved base sequences or regions of those base sequences,

wherein

(e) the set of nucleic acid molecules starting from the nucleic acid molecules that are obtainable or derivable according to steps (a) to (d) were produced synthetically and in at least two separate synthesis batches,

wherein

(f) the individual nucleic acid molecules or some of the nucleic acid molecules hybridise to

(i) different phylogenetically conserved base sequences,  
or

(ii) one and the same phylogenetically conserved base sequence at non-overlapping sequence regions, or

(iii) one and the same phylogenetically conserved base sequence at overlapping sequence regions,

and wherein

(g) the set for an individual nucleic acid molecule, for a number of its individual nucleic acid molecules or for each of its individual nucleic acid molecules in each case comprises at least one further nucleic acid molecule that, in a region of at least 10 successive nucleotides of their nucleotide chains, corresponds to less than 100% but to at least 80% of the base sequence.

2. Set of nucleic acid molecules by means of which, in a process for the detection of representatives of *Salmonella enterica* subsp. *enterica*, *salamae*, *arizonae*, *diarizonae*, *houtenae*, *bongori* and *indica*, all the representatives of those subspecies can be detected, which set of nucleic acid molecules is **obtainable** by

- (a) obtaining or deriving a first nucleic acid molecule (nucleic acid molecule 1) in a manner known *per se* using a nucleic acid isolate of a representative of one of the mentioned *Salmonella enterica* subspecies, which first nucleic acid molecule is specifically suitable as primer or probe for the detection of that representative or of further or all representatives of that one *Salmonella enterica* subspecies and possibly also of representatives of further *Salmonella enterica* subspecies,
- (b) obtaining or deriving a second nucleic acid molecule (nucleic acid molecule 2) in a manner known *per se* using a nucleic acid isolate of a different representative of one of the mentioned *Salmonella enterica* subspecies, which second nucleic acid molecule is specifically suitable as primer or probe for the detection of that representative or of further or all representatives of that different *Salmonella enterica* subspecies and possibly also of representatives of others of the mentioned *Salmonella enterica* subspecies, and
- (c) unless it is already possible to detect all the representatives of the mentioned *Salmonella enterica* subspecies using the nucleic acid molecules obtainable or derivable according to (a) and (b), continuing to obtain or derive nucleic acid molecules according to (a) and/or (b) until all the representatives of the mentioned *Salmonella*

*enterica* subspecies can be detected using the obtained or derived set of nucleic acid molecules, wherein

(d) the nucleic acid isolates comprise or are phylogenetically conserved base sequences or regions of those base sequences,

wherein

(e) the individual nucleic acid molecules or some of the nucleic acid molecules hybridise to

(i) different phylogenetically conserved base sequences, or

(ii) one and the same phylogenetically conserved base sequence at non-overlapping sequence regions, or

(iii) one and the same phylogenetically conserved base sequence at overlapping sequence regions,

wherein

(f) the set for an individual nucleic acid molecule, for a number of its individual nucleic acid molecules or for each of its individual nucleic acid molecules in each case comprises at least one further nucleic acid molecule that, in a region of at least 10 successive nucleotides of their nucleotide chains, corresponds to less than 100% but to at least 80% of the base sequence,

and wherein

(g) the set of nucleic acid molecules does not comprise any degenerate nucleic acid molecules.

3. Set of nucleic acid molecules by means of which, in a process for the detection of representatives of *Salmonella enterica* subsp. *enterica*, *salamae*, *arizonae*, *diarizonae*, *houtenae*, *bongori* and *indica*, all the representatives of

those subspecies can be detected, which set of nucleic acid molecules is **obtainable** by

(a) obtaining or deriving a first nucleic acid molecule (nucleic acid molecule 1) in a manner known *per se* using a nucleic acid isolate of a representative of one of the mentioned *Salmonella enterica* subspecies, which first nucleic acid molecule is specifically suitable as primer or probe for the detection of that representative or of further or all representatives of that one *Salmonella enterica* subspecies and possibly also of representatives of further *Salmonella enterica* subspecies,

(b) obtaining or deriving a second nucleic acid molecule (nucleic acid molecule 2) in a manner known *per se* using a nucleic acid isolate of a different representative of one of the mentioned *Salmonella enterica* subspecies, which second nucleic acid molecule is specifically suitable as primer or probe for the detection of that representative or of further or all representatives of that different *Salmonella enterica* subspecies and possibly also of representatives of others of the mentioned *Salmonella enterica* subspecies, and

(c) unless it is already possible to detect all the representatives of the mentioned *Salmonella enterica* subspecies using the nucleic acid molecules obtainable or derivable according to (a) and (b), continuing to obtain or derive nucleic acid molecules according to (a) and/or (b) until all the representatives of the mentioned *Salmonella enterica* subspecies can be detected using the obtained or derived set of nucleic acid molecules, wherein

(d) the nucleic acid isolates comprise or are phylogenetically conserved base sequences or regions of those base sequences,

wherein

(e) the individual nucleic acid molecules or some of the nucleic acid molecules hybridise to

(i) different phylogenetically conserved base sequences,  
or

(ii) one and the same phylogenetically conserved base sequence at non-overlapping sequence regions, or

(iii) one and the same phylogenetically conserved base sequence at overlapping regions,

wherein

(f) the set for an individual nucleic acid molecule, for a number of its individual nucleic acid molecules or for each of its individual nucleic acid molecules in each case comprises at least one further nucleic acid molecule that, in a region of at least 10 successive nucleotides of their nucleotide chains, corresponds to less than 100% but to at least 80% of the base sequence,

and wherein

(g) the set of nucleic acid molecules comprises only complementing primers.

Q 4. Set according to claim 1, 2 or 3, **characterised** in that the set for an individual nucleic acid molecule, for a number of its individual nucleic acid molecules or for each of its individual nucleic acid molecules in each case comprises at least one further nucleic acid molecule that, in a region of at least 10 successive nucleotides of their nucleotide chains, differs from the other or further nucleic acid molecule in precisely one base position.

Q 5. Set according to ~~any one of the preceding claims~~,  
**characterised** in that it comprises one or more, but not  
exclusively, nucleic acid molecules that are fragments of  
the SEQ ID NO 1 according to WO 95/00 664 or of its  
complementary sequence.

Q 6. Set according to ~~any one of the preceding claims~~,  
**characterised** in that the individual nucleic acid molecules  
hybridise to the same strand of nucleic acid isolates of  
representatives of *Salmonella enterica* subspecies that are  
being subjected to the process for their detection.

Q 7. Nucleic acid molecule that belongs to a set according to  
~~any one of the preceding claims~~ or that can be used for  
such a set, **characterised** in that, in a region of at least  
10 successive nucleotides of its nucleotide chain, the  
sequence of the nucleic acid molecule corresponds exactly  
to a sequence region of at least one representative of  
*Salmonella enterica* subspecies according to ~~any one of~~  
~~claims 1, 2 or 3~~, the sequence region comprising or being a  
phylogenetically conserved base sequence or a region of  
that base sequence.

8. Nucleic acid molecule according to claim 7,  
**characterised** in that, in a region of at least 10  
successive nucleotides of its nucleotide chain, it is 100%  
or at least 80% identical to a corresponding number of  
successive nucleotides of one or more of the following  
sequences or their complementary sequences:

SEQ ID NO: 1 ATGGATCAGAATACGCCCCG

SEQ ID NO: 2 ATGGATCAGAATACACCCCCG

SEQ ID NO: 3 CAGAATACGCCCCGTTTCGGC  
SEQ ID NO: 4 CAGAATACACCCCGTTTCGGC  
SEQ ID NO: 5 CAGAATACGCCCCGTTTCAGC  
SEQ ID NO: 6 CAACCTAACTTCTGCGCCAG  
SEQ ID NO: 7 CAACCTAACTTCTGCACCAG  
SEQ ID NO: 8 CAACCTAACCTCTGCGCCAG  
SEQ ID NO: 9 CAACCTAACTTCTGCGGCAG  
SEQ ID NO: 10 CAGCCTAACTTCTGCGCCAG

9. Nucleic acid molecule **characterised** in that, in respect of its sequence, it is homologous to a nucleic acid molecule according to ~~either claim 7 or claim 8~~ and, in at least 10 successive nucleotides of its nucleotide chain, (i) is identical to a nucleic acid molecule according to ~~either claim 7 or claim 8~~, or (ii) differs from a nucleic acid molecule according to ~~either claim 7 or claim 8~~ in not more than one nucleotide, or (iii) differs from a nucleic acid molecule according to ~~either claim 7 or claim 8~~ in not more than two nucleotides.

10. Nucleic acid molecule according to ~~any one of claims 7 to 9~~, <sup>claim 7</sup> **characterised** in that it is from 10 to 250 nucleotides long, and preferably from 15 to 30 nucleotides long.

11. Nucleic acid molecule according to ~~any one of claims 7 to 10~~, <sup>claim 7</sup> **characterised** in that it is single-stranded or has a complementary strand.



claim 7

12. Nucleic acid molecule according to ~~any one of claims 7 to 11~~, **characterised** in that it is present

- (i) as DNA, or
- (ii) as RNA corresponding to (i), or
- (iii) as PNA, the nucleic acid molecule where appropriate having been modified or labelled in a manner known per se for analytical detection processes, especially detection processes based on hybridisation and/or amplification.

13. Nucleic acid molecule according to claim 12, **characterised** in that it is a modified or labelled nucleic acid molecule in which up to 20% of the nucleotides of at least 10 successive nucleotides of its nucleotide chain are building blocks known per se as probes and/or primers, especially nucleotides that do not occur naturally in bacteria.

14. Nucleic acid molecule according to claim 12 ~~or claim 13~~, **characterised** in that it is a modified or labelled or additionally modified or labelled nucleic acid molecule that comprises, in a manner known per se for analytical detection processes, one or more radioactive groups, coloured groups, fluorescent groups, groups for immobilisation on a solid phase, groups for an indirect or direct reaction, especially for an enzymatic reaction, preferably using antibodies, antigens, enzymes and/or substances having an affinity for enzymes or enzyme complexes, and/or other modifying or modified groups of nucleic-acid-like structure that are known per se.

15. Kit for analytical detection processes, for the detection of bacteria of the *Salmonella* genus,

characterised by

- (i) a set of nucleic acid molecules according to ~~any one~~ <sup>claim 1</sup> of claims 1 to 6, or
- (ii) one or more nucleic acid molecules according to ~~any one of claims 7 to 14~~ <sup>claim 7</sup>.

16. Kit according to claim 15, characterised in that the set of nucleic acid molecules was produced synthetically and that it was produced in at least two separate synthesis batches.

17. Kit according to claim 16, characterised in that the kit does not comprise any degenerate nucleic acid molecules.

18. Use of a set of nucleic acid molecules according to ~~any one of claims 1 to 6~~ <sup>claim 7</sup>, of one or more nucleic acid molecules according to ~~any one of claims 7 to 14~~ <sup>claim 7</sup> or of a kit according to ~~any one of claims 15 to 17~~ <sup>claim 15</sup> to detect the presence or absence of bacteria belonging to representatives of *Salmonella enterica* subspecies according to claim 1, 2 or 3.

19. Use according to claim 18, characterised in that nucleic acid hybridisation and/or nucleic acid amplification is carried out.

20. Use according to claim 19, characterised in that a polymerase chain reaction (PCR) is carried out as nucleic acid amplification.

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21. Use according to claim 18, ~~19 or 20~~, characterised in that differences between the genomic DNA and/or RNA of the bacteria to be detected and of the bacteria that are not to be detected are determined at at least one nucleotide position in the region of a nucleic acid molecule according to ~~any one of claims 7 to 14~~ <sup>claim 7</sup> and representatives of a group of bacteria of the *Salmonella* genus are detected, especially representatives of *Salmonella enterica* subspecies according to claim 1, ~~2 or 3~~.

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